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MYELINATED AFFERENT FIBRES RESPONDING SPECIFICALLY TO NOXIOUS STIMULATION OF THE SKIN

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SUMMARY

1. The characteristics of receptors from the hairy skin of the hind limb of cat were studied by recording from single primary afferent fibres with fine micropipettes. The distinctive features of 513 fibres conducting under 51 m/sec are described.

2. Seventy-four fibres conducting between 6 and 37 m/sec were classified as nociceptors because they responded only to damaging mechanical stimulation of the skin. These fibres responded maximally to pinching the skin with a serrated forceps or to cutting the skin. Noxious heat, noxious cold, acid applied to the receptive field and bradykinin injected into skin cuts did not evoke discharges from such receptors. Typically their receptive fields were 2–5 cm long by 1–2·5 cm wide and consisted of responsive spots (under 1 mm diameter) separated by unresponsive areas. There was a tendency for the most slowly conducting fibres so classified to be the least sensitive.

3. Other afferent fibres had receptive fields similar to the nociceptors; however, they were excited by substantial but not noxious mechanical deformation. Their conduction velocities overlapped those of the nociceptors and extended upwards to 51 m/sec; the most rapidly conducting fibres tended to be the most sensitive to mechanical stimuli. These insensitive mechanoreceptors or moderate pressure receptors adapted more slowly than the nociceptors.

4. The majority of fine myelinated axons originated from hair receptors and had conduction velocities concentrated between 14 and 22 m/sec.

5. The possible relation of these observations to pain and reactions typical of pain is considered.

INTRODUCTION

Despite considerable study of cutaneous sense organs, the relation between receptors and pain remains uncertain. This is particularly surprising for receptors with myelinated axons, since evidence exists that pain arises from activity in myelinated afferent fibres, and activity in such fibres is relatively easy to study.

In man, reaction time measurements show that after a noxious stimulus, pain is felt too quickly to have been mediated exclusively by unmyelinated fibres (Zotterman, 1933; Lewis & Pochin, 1937; Gordon & Whitteridge, 1943). Experiments employing electrical stimulation of cutaneous nerves in both man and dog have demonstrated that pain and associated reactions occur whenever the smallest myelinated fibres are stimulated (Heinbecker, Bishop & O'Leary, 1933; Bishop & Heinbecker, 1935; Collins, Nulsen & Randt, 1960). However, only a few myelinated afferent fibres excited specifically by noxious stimulation of the skin have been found (Maruhashi, Mizaguchi & Tasaki, 1952; Hunt & McIntyre, 1960) and this fact has supported theories attributing pain to some characteristic temporal-spatial pattern of activity in a population of cutaneous receptors which also respond to non-damaging stimulation (see Melzack & Wall, 1965). The need for further information concerning the way in which receptors associated with small myelinated fibres respond to noxious stimuli motivated the present study. It is relatively novel in approach since micro-electrodes were used to record from single fibres in cutaneous nerve (Tasaki, 1952; Miller, Taylor & Weddell, 1962) allowing analyses of a large number of afferent fibres in a particular experiment.

METHODS

Young adult cats (2–3 kg) were anaesthetized with sodium pentobarbital (Nembutal, Abbott Laboratories) and given regular supplementary doses sufficient to maintain complete areflexia. The animal was rigidly supported at the hips in the prone position with the left hind limb extended. Rectal temperature was maintained at 37·5° C (±0·2° C). Room temperature was held at 30° C and external radiant heat was used to keep skin and oil pools temperatures at about 36·5° C. The general condition of the animal was monitored by recording the systemic arterial pressure and end-tidal CO_2.

All but two of the thirty-six experiments were done on the posterior femoral cutaneous nerve, which innervates the dorsal skin of the hind limb from the proximal thigh to the ankle. The nerve was exposed near the hip and partially freed from the surrounding connective tissue by dissection under a binocular microscope, leaving the blood supply to the nerve largely undisturbed. Figure 1 shows the usual experimental arrangement. Stimulating electrodes (*A*) were placed centrally on the nerve for activation of fibres and measurements of conduction velocity. In five experiments, a laminectomy was performed so that stimulating electrodes could be placed on dorsal root fibres (L-7 and S-1) for comparison with conduction velocity measurements obtained by direct stimulation of the nerve. The nerve

was covered with liquid paraffin and a platform (*B*) was placed under the nerve lifting it from the surrounding tissue near the distal edge of an oil pool formed with cut skin. A gross recording electrode (*C*) was located just distal to the platform to monitor the compound action potential. Glass micropipettes filled with 4 M-NaCl, 20–80 MΩ in impedance (at 1000 c/s, held in a microdrive, were used to penetrate that portion of the nerve lying on the platform. Action potentials recorded between an indifferent electrode (hip pin) and the micro-electrode were led to a high input-impedance, unity-gain transistorized amplifier with

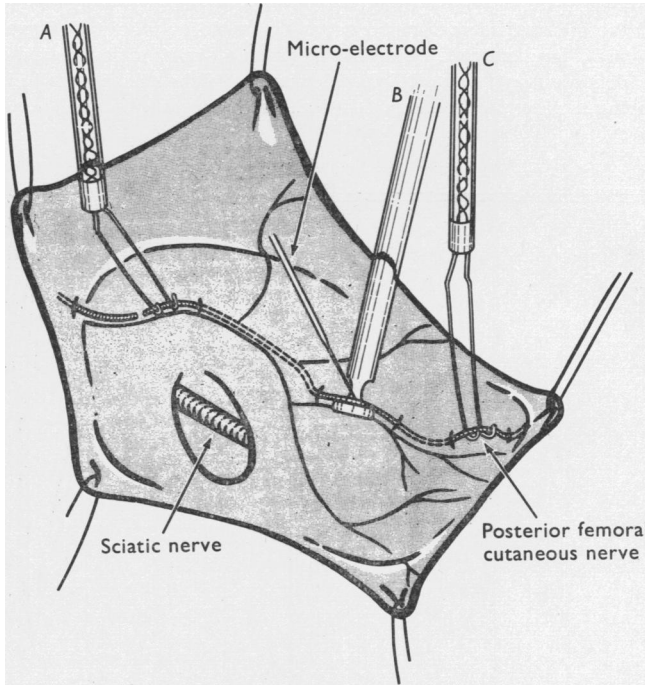

Fig. 1. Experimental arrangement used for micro-electrode recordings from afferent fibres of the posterior femoral cutaneous nerve. The shaded area indicates the oil pool. *A*, the stimulating electrode; *B*, the platform; *C*, the electrode for recording the gross potential.

negative capacity feed-back. The output of this impedance-matching device was amplified and filtered to enhance the signal-to-noise ratio, displayed on an oscilloscope, reproduced through an auditory monitor, and made available at the input of a magnetic tape recording system employing FM electronics. The normal recording bandpass was from approximately 100 to 3000 c/s. This produced some alteration of spike configuration but no significant effect upon latency. Conduction velocity was calculated from response latencies as photographed from an oscilloscope at threshold and at thrice threshold for a particular fibre, and from a measurement of conduction distance made *in situ* at the termination of the experiment. (A fine thread was laid along the course of the nerve from the stimulating cathode to the recording point on the Perspex platform, the thread being arranged to follow every portion of the nerve path.) Time for stimulus utilization at the stimulating electrode was not taken into account. Only slight shifts in latency (0·05–0·2 msec) accompanied changes in stimulus intensity from threshold for a particular fibre to 5 times threshold as long as

there was no evidence of conduction block. For the fibres which shall be considered, the latency shifts between threshold and thrice threshold changed the calculated conduction velocities by no more than 3 m/sec. In five experiments conduction velocity as calculated from the latency of a response evoked from the nerve stimulating electrode was compared to that calculated from the response latency to dorsal root stimulation; no significant differences (greater than 3 m/sec) were found.

Parameters of the stimulus and stimulus markers were recorded in parallel with neural discharge on different channels of the magnetic tape. The magnetic tape records were analysed by replaying them into a high frequency galvanometer oscillograph (bandpass flat to 3000 c/s), one channel of the oscillograph recording the neural discharge, another the voltage proportional to the instantaneous frequency of the discharge (from a digital time-interval counter with storage and analogue output) and a third, the measure of the stimulus. All illustrations of impulses occurring in response to adequate stimuli were made from oscillograph records.

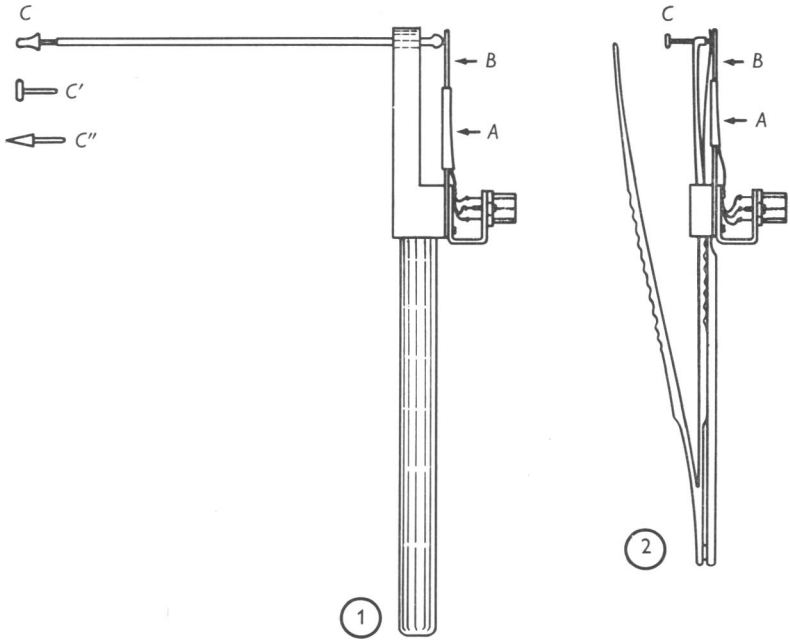

Fig. 2. Stimulators used for applying pressure to the skin. Stimulator 1 was used to apply force perpendicular to the skin while stimulators such as 2 were used to pinch the skin. For both stimulators, *C* was mounted on a plunger which passed freely through a guide to press against a phosphor-bronze strip, *B*, supported at one end. Deflexion of *B* was sensed by a strain gauge, *A*. *C'*, *C''* are different tips used with stimulator 1.

A variety of stimulators were available including the two types shown in Fig. 2, which were used to deliver calibrated mechanical stimuli. The devices of Fig. 2 had a plunger which deflected a fine alloy strip (*B*) on which a strain gauge was mounted (*A*). The strain gauge output was displayed on the oscilloscope and recorded on magnetic tape. The amount of force required for given degrees of strain gauge output was determined using a balance. In addition, calibrated von Frey hairs mounted on wooden rods, a glass rod with a fire-polished tip 1·5 mm in diameter, different types of forceps, and several devices for radiant

or direct contact heating of the skin were employed for stimulation. Cooling was accomplished by evaporation of ether or ethyl chloride from the skin. A method of stimulation which was used to classify mechanoreceptors and, in particular, to deliver a constant forcible stimulus, consisted of grasping a fold of skin with a clip equipped with a flat, padded bill which exerted a force of 700 g. (The contact area of the clip was approximately 4 by 5 mm and when it was used on the skin of the human thigh gave rise to an incrementing painful sensation. Some alert and ambulatory cats obviously were disturbed by the presence of this clip when it was applied to the posterior thigh skin, while in others it did not produce any overt reaction.) Some experiments were done with the hair uncut upon the skin and a few with the hair clipped to 2–3 mm in length. In the majority of the experiments hair was removed from the skin by a chemical depilatory. The skin area stimulated could be observed through a binocular dissecting microscope at magnifications between 6 and 25×. Other details pertaining to stimulation are described with the results.

RESULTS

When high impedance (20–80 MΩ) micro-electrodes were inserted into the nerve and slowly advanced while the nerve was electrically stimulated (2/sec) at an intensity maximal for all myelinated fibres, a small field potential was usually recorded which corresponded to the large deflexions of the compound action potential. From time to time initially positive deflexions appeared, like those in Fig. 3. Reduction of stimulus intensity showed that such positive-going potentials disappeared in an all-or-none fashion at a given stimulus intensity and therefore originated from a single fibre. The action potentials were only rarely superimposed upon a typical transmembrane potential, indicating that they were usually obtained from an extracellular location.

Unitary potentials recorded in this manner were typically large, ranging from 2 to 30 mV. The stability of the recording for the more rapidly conducting fibres was generally excellent and it was possible to 'hold' units for long periods of time. With fibres conducting under 30 m/sec the stability of recording varied and some were lost before studies were complete. Fibres on which insufficient data were obtained for identification of the nature of the receptor are not included in the population described.

In a typical experiment with the nerve in good condition throughout the recording period (up to 18 hr), more than 98 % of the units studied could be identified by the receptor type. The method sampled fibres conducting from 6 to 80 m/sec, and seemed to encompass the entire myelinated spectrum. Fibres conducting over 40 m/sec were seen more commonly than more slowly conducting elements, even though small-diameter myelinated fibres are numerous in the posterior femoral cutaneous nerve. Attention was focused upon fibres conducting under 35 m/sec, although ordinarily a cursory examination of receptive characteristics was made for every fibre encountered.

The latency of the unitary response to electrical stimulation of the nerve,

the location of the receptive field, and the general characteristics of the response to adequate stimulation were used to distinguish a given fibre and to determine whether it had been previously observed. Without deliberate movement of the micro-electrode the recording rarely shifted so that action potentials were first obtained from one and then another fibre.

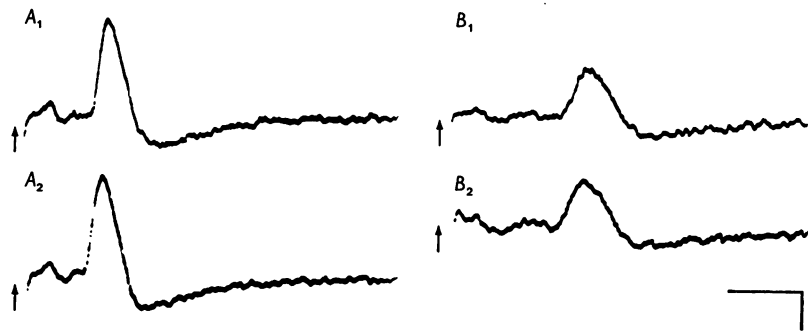

Fig. 3. Unitary responses to electrical stimulation from the posterior femoral cutaneous nerve A_1, B_1, show response for two fibres at threshold stimulation and A_2, B_2 the response of the same fibres at 3 × threshold. Conduction distance for all responses was 21 mm. Calculated conduction velocity for A fibre was 23·4 and for the B fibre 13·5 m/sec. Arrows indicate time of stimulus. Vertical calibration, 5 mV. Horizontal calibration, 1 msec.

Five hundred and thirteen fibres conducting under 51 m/sec make up the subject of this report. The great majority of these units were studied to the point that further investigation was deemed unnecessary. Typically, observations in even the more slowly conducting units were continued for 15–20 min and many units were studied for more than an hour. Thus, the method, at least as far as myelinated fibres are concerned, is an efficient though not necessarily ideal technique for isolating one fibre's activity from that of others.

Resting or background activity was rare in this population of afferent fibres, and absent in fibres conducting under 30 m/sec (437 examples). A very few fibres conducting between 30 and 50 m/sec exhibited some background discharge, particularly after stimulation (see below). All the more rapidly conducting fibres were confirmed to originate from sensitive mechanoreceptors of one type or another (Witt & Hensel, 1959; Hunt & McIntyre, 1960; Iggo, 1965).

Nociceptive fibres

It was apparent in the first experiments that a number of fibres conducting under 35 m/sec required intense mechanical stimuli for excitation. They regularly responded when a forceps with teeth (serrated) was used

to pinch a specific area of the skin. Various innocuous forms of mechanical stimulation were ineffectual when applied to the same area, as was sudden cooling to below 20° C or heating the skin to well above 50° C. An example of the response from this type of unit is shown in the upper traces of Fig. 4*A* and *B*. In Fig. 4*A*, the lower of the pair of traces shows the strain

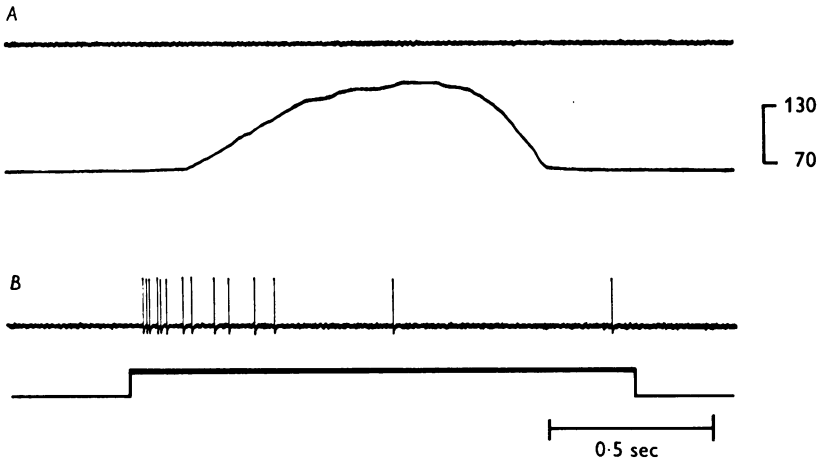

Fig. 4. Responses of a nociceptive fibre to mechanical stimuli. Fibre conduction velocity 25 m/sec. Upper traces in *A* and *B* show action potentials. *A*. Stimulus was delivered by device of Fig. 2-2 to a fold of skin. Force is shown on lower trace by strain gauge output. Calibration on right in g. *B*. Fold of skin in same area firmly squeezed by serrated forceps for duration of upward deflexion on lower trace.

gauge output of the forceps stimulator illustrated in Fig. 2-2. Squeezing any portion of the receptive field with the forceps stimulator fitted with a 2·1 mm tip so that over 130 g of force was exerted across a fold of skin did not elicit a response (upper trace of Fig. 4*A*). Yet, grasping the skin with the serrated forceps evoked activity from a number of points as in Fig. 4*B*.

Figure 5 shows the response of two other units with mechanical thresholds comparable to that of the type illustrated by Fig. 4. For Fig. 5*A* the plunger-stimulator of Fig. 2-1 with a sharp (needle) point (*C″*) at its end was used to explore the skin systematically while recording from a fibre with a conduction velocity of 13·5 m/sec. The force exerted with the sharpened probe is indicated on the lower trace for three successive stimulations of different but nearby points (1–2 mm apart) in the centre of the receptive field. As Fig. 5*A* shows, one stimulation did not evoke a response despite the fact that the force exerted by the needle exceeded 130 g; two other stimulations provoked a short burst of activity only when the force was well in excess of 70 g. Such stimuli were sufficient to drive the needle

partway through the epidermis and left visible holes. In some fibres such as the one illustrated in Fig. 5*B*, even forcible stimulation with a needle did not evoke a consistent response. Yet, activity was regularly initiated by squeezing the skin with the serrated forceps firmly enough to cause obvious damage. Typically, as in Fig. 5, the fibres with the highest threshold to mechanical stimuli had conduction velocities under 20 m/sec, although some conducted more rapidly.

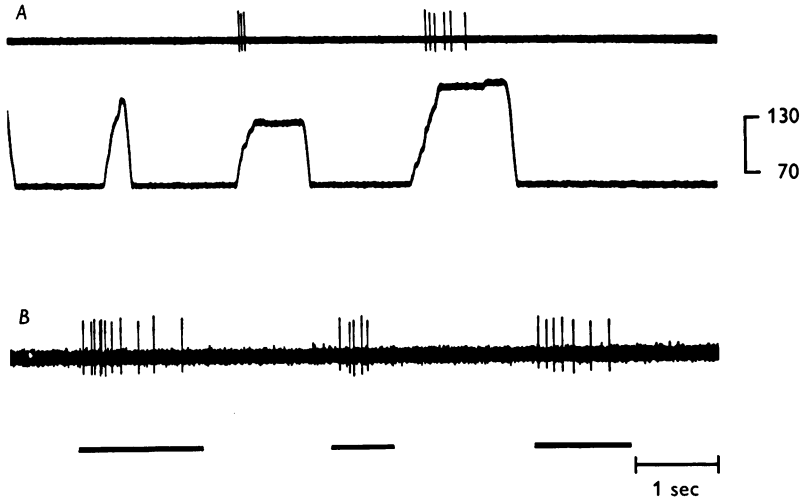

Fig. 5. Response of two nociceptive fibres to mechanical stimuli. Both fibres are from the same experiment. *A*. Same fibre as Fig. 3-*B*. Receptive field shown in Fig. 7*D*. Stimulator of Fig. 2-1 used to push needle (*C''*) against closely approximated points in centre of field. Force is shown on lower trace by strain gauge output. Calibration in g. *B*. Fibre conduction velocity, 9·5 m/sec. Receptive field shown in Fig. 7*A*. Bars indicate approximate period of pinching the skin in the centre of the receptive field by serrated forceps.

Figure 6 illustrates the response of a receptor somewhat more sensitive than the three just described. As shown in Fig. 6*A*, this fibre did not respond to the forceps with the blunt (2·1 mm) tip when a fold of skin was pinched with a force in excess of 100 g. On the other hand, the unit responded well to the needle stimulator (Fig. 6*B*) and gave a high frequency discharge of short duration when the skin was grasped by the serrated forceps (Fig. 6*C*).

Fibres such as those illustrated in Figs. 4 and 5 gave no response when the clip described in Methods was applied to the receptive area mapped by a needle or serrated forceps. On the other hand, the fibre illustrated in Fig. 6 and a number of other nociceptors were excited by this clip. The response to the clip in Fig. 6*D* shows a typical feature: the discharge

increased and then decreased; yet the pressure on the fold of skin exerted by the clip was constant.

Fibres similar to those of Figs. 4–6 were excited to their maximal discharge frequency when small cuts were made in the skin, the response occurring only during the process of cutting. The shortest interval between discharges in an evoked burst from a cut ranged between 5 and 10 msec.

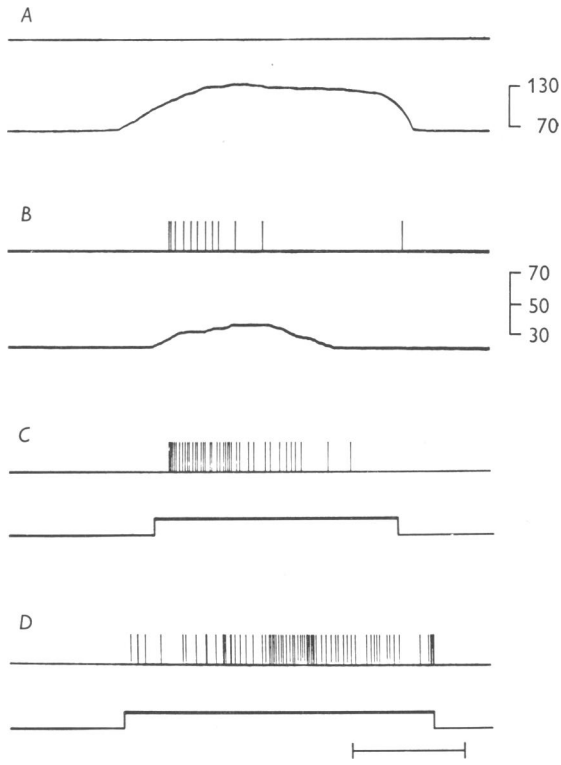

Fig. 6. Responses of a more sensitive nociceptive fibre to mechanical stimuli. Fibre conduction velocity, 29 m/sec. Receptive field shown in Fig. 7*E*. Upper traces in *A*, *B*, *C* and *D* show the output of a pulse circuit triggered by the unitary action potentials. *A*. Device of Fig. 2-2 used to stimulate responsive point. *B*. Device of Fig. 2-1 with needle used as stimulator. *C*. Fold of skin in the centre of the receptive field squeezed with serrated forceps for approximate duration of upward deflexion in lower trace. *D*. Clip (see methods) used to pinch the centre of the receptive field for approximate duration of upward deflexion in lower trace. Calibrations on right in g. Time marker equivalent to 0·5 sec for *A*, *B*, *C* and 8 sec for *D*.

These same receptors were almost completely insensitive to noxious heat, giving rise to no more than a rare impulse even when the skin was charred. The insensitivity to chemical stimuli was another striking feature. Strong acid or bradykinin applied to abraded skin evoked little or no response.

In addition, these fibres showed no increase in sensitivity as a result of repeated stimulation. Rather, repeated stimulation of a given point in the receptive field by the serrated forceps or the needle resulted in decreased responsiveness so that eventually no discharge could be evoked from that particular spot. Quantitative studies of the recovery of excitability after deactivation were not feasible for these insensitive elements, although it was clear that after 1 or 2 min there was often some return of responsiveness, while after lesser waiting periods the fibre was still inexcitable. If a given spot was rendered insensitive by repeated stimulation, no detectable difference in the response from other spots was observed. Units which required forcible pinching of the skin with the toothed forceps for excitation were sometimes rendered inexcitable by a single stimulation.

Velocity of stimulation did influence the response of these mechanoreceptors, but it was not a crucial factor. Thus, in Fig. 4*A*, the highest discharge frequency occurred during the period of rapid change in the stimulus, while in Fig. 6*D* the maximal discharge frequency took place well after the constant (clip) stimulus had begun. The small area of the sensitive spots, the noxious quality of the adequate stimulus and the inactivation by repeated stimuli made quantitative studies of velocity sensitivity impossible. As Figs. 4–6 illustrate, the nociceptive fibres tended to adapt moderately rapidly to the adequate stimulus. The higher the mechanical threshold, the more rapid was the adaptation rate; however, it should be noted that the stimuli caused overt damage.

A total of seventy-four fibres from the posterior femoral cutaneous nerve were classified as nociceptive mechanoreceptors. The minimal adequate stimulus in every case ranged from one which was frankly noxious, to the clip used in Fig. 6*C*, which was merely painful when applied to human skin. The existence of afferent fibres with essentially the same characteristics in other cutaneous nerves was readily confirmed in experiments on the sural and plantar nerves. In one experiment, a 19 m/sec fibre of the nociceptive class from the posterior femoral cutaneous nerve was extensively studied in a filament dissected from the S-1 dorsal root.

Receptive fields of nociceptive fibres. The nociceptive terminals were located in the skin since the receptive points moved with movement of the skin relative to more deeply located tissue. Furthermore, one of the most effective stimuli was pinching a fold of skin lifted from the underlying fascia and muscle. The area of skin from which responses could be evoked was systematically mapped for a number of the nociceptors. Whenever possible, mapping was done using the needle (*C″*) mounted on the stimulator of Fig. 2-1. Positive responses, as judged from the oscilloscope and the auditory monitor, were marked with washable ink. When the mapping

was completed, the receptive field was photographed. The range of sizes of receptive fields for nociceptive mechanoreceptors is shown in Fig. 7*A–E*. Typically, the receptive fields were large, extending over 2 cm in the distal-proximal direction and over 1 cm in the medial-lateral direction. In some

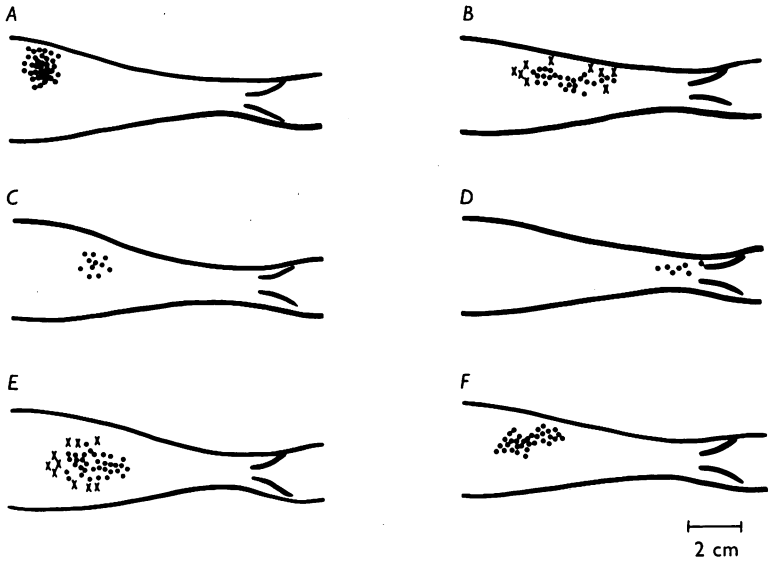

Fig. 7. Receptive fields: five nociceptors (*A–E*) and one insensitive mechanoreceptor (*F*). The receptive fields were traced from photographic enlargements made to same scale and projected on an outline drawing of the dorsum of the cat's leg from the hip to ankle. Points marked in receptive field are accurately located in terms of position, one to another, but their size does not indicate size of responsive spots. *B* and *E* have × 's to mark extension of field plotted by needle stimulation, when serrated forceps were employed. Conduction velocity *A*, 9·5 m/sec; *B*, 26·8 m/sec; *C*, 25·2 m/sec; *D*, 13·5 m/sec; *E*, 29·0 m/sec; *F*, 23·4 m/sec.

instances, they covered much of the distribution of the nerve; the latter could be approximated by listening to the recording from the gross electrode while stroking the skin. Figure 7*A* shows a representative receptive field, while Fig. 7*B* illustrates the largest receptive field found for the nociceptive class. Figure 7*C* and *D* give two of the smallest receptive fields observed for the group.

The spots as shown in these illustrations indicate particular sensitive points from which responses were evoked while the absence of a marked area between spots indicates a silent region. The size of the spots on the reproductions is not an accurate indication of the size of the responsive region. The noteworthy observation in the mapping experiments was the distinctly distributed nature of the receptive fields. Furthermore, the shape of most receptive fields made it unreasonable to believe that the stimuli

were directly exciting an axon or major axon branches. There was no clear correlation between the size of the receptive field and the conduction velocity of the fibre; however, fibres conducting particularly slowly, those under 20 m/sec, were frequently very insensitive and the receptive fields for many of these fibres were smaller than those for more rapidly conducting fibres. This may simply have been due to the intensity of stimulus necessary to excite the receptor with only the more sensitive spots being marked. Occasionally, a systematic attempt was made to alter a field by using the maximal possible stimulus intensity with the serrated forceps and some fields enlarged as Fig. 7*B* and *E* (×), while many were unchanged.

Mechanical threshold. As the above description indicates, the sensitivity to mechanical stimuli was low. All but one of those tested did not respond to a 3·3 g von Frey hair at any spot in the receptive field. In several instances, special von Frey hairs exerting 5–8 g were prepared and with these a response could be obtained from the most sensitive points in the receptive field. It was noted that such extremely stiff hairs penetrated the skin treated with the depilatory, and thus were considered noxious. When a non-penetrating stimulus was used, some of the fibres classified as nociceptive, as the one of Fig. 6 and 7*E*, gave a response, provided that the skin was pinched with sufficient force (the clip of Fig. 6*D*). A calibrated tweezer of the type illustrated in Fig. 2-2, capable of exerting high force, was used in certain experiments to test the sensitivity of the receptors with the very highest mechanical thresholds. A few discharges were obtained when the skin was pinched by forces between 300 and 500 g; these stimuli raised obvious welts and inactivated the skin region for many minutes. At the most sensitive points in the receptive field nociceptors responded to a sharp point when 10–100 g of force was used to press against the skin. Other mechanoreceptors such as 'touch spots' or 'domes' (Hunt & McIntyre, 1960; Iggo, 1963) responded well to a 0·025 g von Frey hair.

Conduction velocity. The histogram in Fig. 8*A* illustrates the frequency of occurrence of receptors classified as nociceptive as a function of conduction velocity. The seventy-four fibres so classified conducted between 6 and 36 m/sec. These fibres represented approximately 15% of the fibres conducting under 30 m/sec located in the study. The mean conduction velocity for the group was 19·5 m/sec; over one half conducted under 19 m/sec. Nociceptive receptors were encountered in 90% of all of the experiments done and as many as seven were studied in a single experiment. The units with the very highest thresholds tended to be the most slowly conducting (under 15 m/sec). With few exceptions, the nociceptive fibres conducted at velocities either just below or just above that equivalent to the main 'delta' peak in the compound action potential of the nerve.

Other insensitive mechanoreceptors with slowly conducting afferent fibres. Other insensitive mechanoreceptors were encountered in these experiments which were similar in some respects to the nociceptive mechanoreceptors just described. An important feature in common between the nociceptors and these more sensitive elements was the nature of the receptive field.

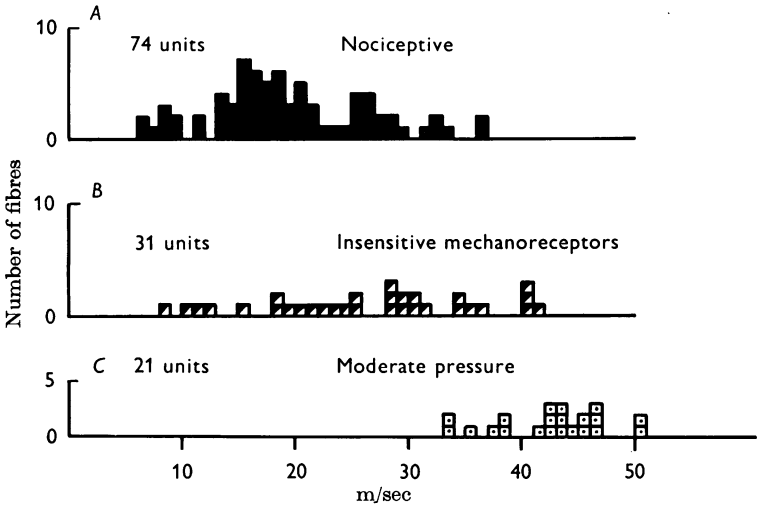

Fig. 8. Plot of frequency of occurrence of afferent fibres as a function of conduction velocity. *A*. Fibres classified as nociceptive from all experiments in series. *B*. Fibres classified as 'insensitive mechanoreceptors' from all experiments. *C*. The more sensitive 'moderate-pressure receptors' from the last eight experiments (see text).

Figure 7*F* illustrates the receptive field of a fibre conducting at 23 m/sec which responded to strong but not frankly noxious mechanical stimuli. The receptive field organization is evidently similar to that of the nociceptors. Receptors were considered to fall into the category of insensitive mechanoreceptors if they did not consistently respond to blunt pressure (Fig. 2-1, tip *C*) of up to 100 g applied at right angles to the skin, but were regularly activated by a forcible pinch with a blunt forceps (Fig. 2-2). The responses of a fibre conducting at 28 m/sec are shown in Fig. 9; it did not respond to vigorous stroking of the skin with the glass rod. Also, no response could be initiated by a smooth disk pressed against the skin with over 80 g of force (Fig. 9*A*). When the same region of the receptive field, however, was grasped by the blunt forceps, certain spots yielded activity (Fig. 9*B*). These fibres as a group responded at highest frequencies to the serrated forceps and to a needle. They had von Frey hair thresholds of from 0·5 to 3·3 g, approaching the nociceptive group at the upper limit. In our sample they were less than one-half as common as the fibres classified as nociceptive. A histogram of the frequency of their

occurrence as a function of conduction velocity is shown in Fig. 8*B*; the mean conduction velocity was 26·4 m/sec and one half conducted over 28 m/sec. At the same time, the slowest fibres completely overlapped the nociceptive units. The responsive characteristics of these insensitive fibres were also quite similar to those of the nociceptive class in that they gave a

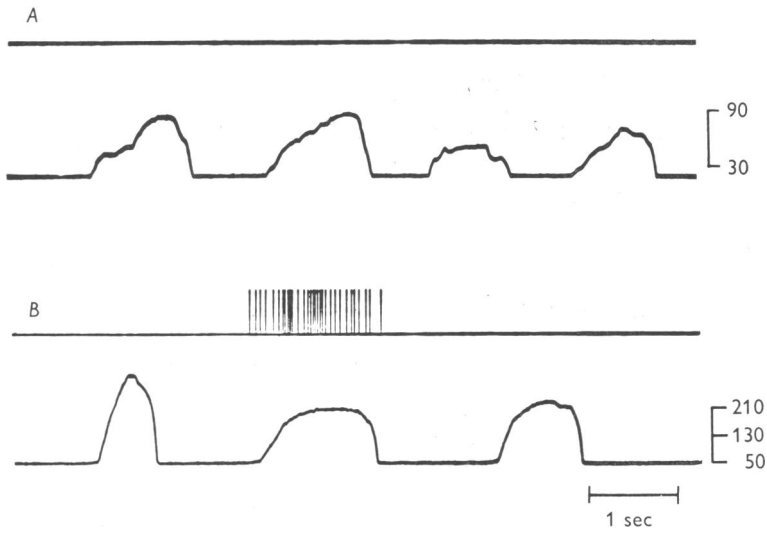

Fig. 9. Response of an insensitive mechanoreceptor with axon conducting at 28 m/sec. Potentials in upper trace of each pair derived as in Fig. 6. Lower traces are stimulator outputs. *A*. Stimulation of centre of receptive fields with device of Fig. 2-1 using the blunt tip (*C*). *B*. Pinch to centre of receptive field with device of Fig. 2-2. Calibrations on right in g.

brief, relatively high-frequency discharge to the serrated forceps. They were inactivated by strong stimulation in much the same way as the nociceptive group, though generally they adapted more slowly than the nociceptors to constant stimulation with a needle. All the fibres indicated in Fig. 8*B* responded at significantly higher frequency to a sharp than to a blunt stimulator, regardless of the pressure exerted by the latter. It must be kept in mind that the mechanical sensitivity of these 'insensitive' mechanoreceptors was usually determined in skin from which hair was removed and with the hair present, blunt stimuli were even less effective.

Moderate-pressure receptors

In the course of this study, a number of afferent fibres were encountered whose receptive fields also were similar to the nociceptors, but which differed in other characteristics. Once again the receptive fields consisted of points of heightened sensitivity distributed over a skin area of from

2 to 3 cm in the rostral-caudal dimension and 1½ to 2 cm in the medial-lateral direction. Though the sensitivity of the receptors in this group varied, their threshold to mechanical stimuli was distinctly lower than either class mentioned above. Most responded with a few discharges to light stroking of the skin with a blunt glass rod, and rubbing the skin more vigorously with the glass rod often gave rise to a relatively high-frequency discharge. Some units, while not responding to gentle stimulation with the glass rod, responded well to the blunt stimulator shown in Fig. 2-1, but maximal discharge for the group was ordinarily produced by stimulation with a needle. Discharge frequency was typically higher for these receptors than for the nociceptors or insensitive mechanoreceptors and occasionally intervals as short as 2–3 msec were observed. In addition, discharge was more slowly adapting than for the less sensitive receptors, some discharge being maintained for many seconds when the blunt stimulator was used. With moderate pressure against the skin or with a non-noxious pinch of the skin with the stimulator of Fig. 2-2, a regular pattern of activity was frequently recorded. Such units also tended to show some after discharge when the effective stimulus was strong or when the skin was stroked with either a needle or repeatedly stroked with a blunt stimulator. A few exhibited a weak discharge to rapid cooling of the skin. The von Frey hair thresholds of such 'pressure' receptors ranged from 0·1 to 0·8 g. At their upper limit the von Frey hair thresholds overlapped sensitivity with the 'insensitive' receptors. In common with some of the 'insensitive' receptors, the 'pressure' receptors sometimes showed an incrementing discharge to moderately strong and steady stimulation. The conduction velocities of twenty-one such units from eight experiments are shown in Fig. 8*C*. The frequency of occurrence is not comparable with Fig. 8*A*, since these units were systematically examined only late in the study. In the last six experiments of the series they and nociceptors were equally common. The mean conduction velocity of the twenty-one fibres in Fig. 8*C* was 42·5 m/sec.

Hair receptors with slowly conducting myelinated afferent fibres

The majority of fibres (78%) conducting above 6 m/sec and under 30 m/sec originated from hair receptors, confirming previous reports (Hunt & McIntyre, 1960; Iggo, 1965). Such fibres were driven to maximal discharge by displacement of the hair. Discharge frequency did not increase when the skin was also stimulated. Adaptation to constant pressure or hair deflexion was the most rapid of all the afferent fibres studied and receptors of this type did not discharge in the absence of movement. On the average, hair receptive fields were much smaller than any of the fields described above and relatively uniform in sensitivity. With the fur removed

from the skin by a depilatory, the situation most common in our experiments, the force of an 0·008 g von Frey hair readily excited just one class of receptors with slowly conducting myelinated fibres. Hair receptors were recognized in the depilated skin by their extreme sensitivity, rapid adaptation and features of the receptive field.

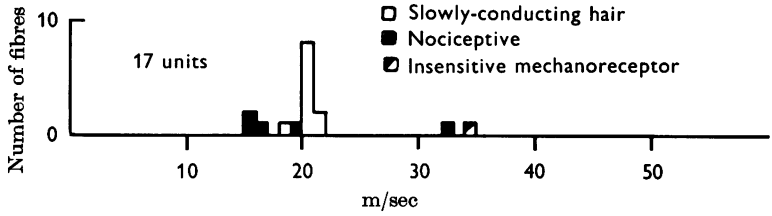

Fig. 10. Classification of all fibres with slowly conducting axons from one experiment.

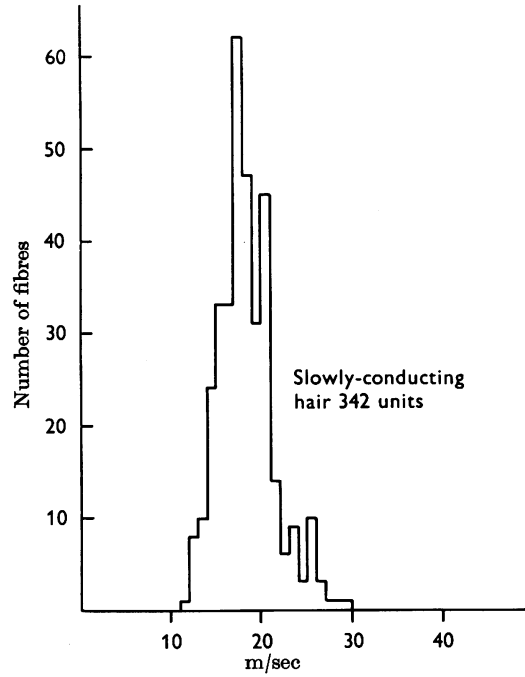

Fig. 11. Conduction velocities of axons from mechanoreceptors of the type maximally stimulated by hair deflexion. Observations from all experiments (34) in series. Mean value: 18·3 m/sec.

Another characteristic of the fibres from the hair receptors in a given experiment was a remarkable uniformity in conduction velocity. Figure 10 illustrates the slowly conducting axons encountered in one experiment. Those classified as hair receptors are shown by open squares; note the congregation of conduction velocities in a very narrow range. In contrast,

the conduction velocities of the nociceptive units in this experiment varied from 14 to 32 m/sec while one unit classified as 'insensitive' had a conduction velocity of 33 m/sec. The limited range of conduction velocities for hair receptors with slowly conducting axons was so striking that, if a

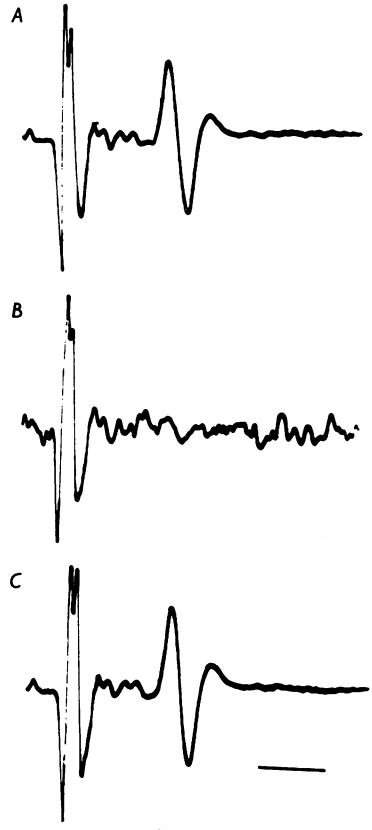

Fig. 12. Compound action potential from posterior femoral cutaneous nerve and the effects of stimulation with an air jet. Action potential evoked at 5/sec by single shocks from a proximally located electrode at the beginning of oscilloscope sweep. Hair on limb left intact. Recording as in Fig. 1 with distally located, gross electrode. *A*. Control. *B*. During movement of airstream across receptive field. *C*. Immediately after airstream stimulation ceased. Time calibration, 1 msec. Conduction distance, 37 mm.

fibre whose latency fell within the main delta deflexion was observed, in nine out of ten cases it would have a mechanical threshold typical of hair receptors. Figure 11 shows a histogram of frequency of occurrence as a function of conduction velocity for afferent fibres from the hair receptors studied in the present series. Much of the spread in distribution illustrated in Fig. 11 results from variation of the modal conduction velocity in different animals.

The results of the single fibre analysis on the relation between conduction velocity and receptor type were supported by two experiments using the collision technique of Douglas & Ritchie (1957), a method which employs changes in electrically initiated compound action potentials to indicate populations of fibres excited by natural stimulation. In these experiments, the hair over the distribution of the posterior femoral cutaneous nerve was left uncut. The compound action potential for all myelinated fibres was recorded from the nerve between the stimulating electrode and the region of the skin served by the nerve. A stream of air sufficient to deflect the hairs was then moved across the skin so that the entire receptive region was stimulated. As a consequence of receptor excitation, collision and/or refractoriness in fibres between the stimulating and recording electrodes would block their contribution to the electrically evoked compound potential. As shown in Fig. 12*B*, such stimulation essentially occluded the 'delta' peak. Both collision experiments gave identical results and were interpreted as indicating that almost all of the receptors served by fibres comprising the 'delta' peak could be excited by hair movement since, of the afferent fibres conducting under 30 m/sec, only these were excited by an air stream. The nociceptors and insensitive mechanoreceptors had both more rapidly and slowly conducting afferent fibres and contributed to the small deflexions on either side of the delta peak (Fig. 12*A*).

DISCUSSION

In contrast to earlier efforts, the present experiments were successful in defining a significant population of cutaneous receptors activated exclusively by noxious mechanical stimuli probably because we were able to sample the activity of a large number of slowly conducting myelinated fibres. The observations, however, have considerable precedent. The early experiments of Adrian (1931) on frog skin implicated small fibres in the responses to a variety of noxious stimuli. Subsequently, work on mammals also related discharges in small cutaneous afferent fibres to damaging stimuli (Zotterman, 1936, 1939; Boman, 1954). Hunt & McIntyre (1960) in their survey of sural nerve afferent fibres described seven out of a total of 421 myelinated fibres which apparently had properties, including conduction velocity, comparable to the nociceptors and insensitive mechanoreceptors described above. It is also possible that Maruhashi *et al.* (1952) observed similar receptors, although they noted a response to heating as well as mechanical stimuli. In addition, it seems pertinent that many afferent fibres from muscle conducting under 25 m/sec originate from high-threshold pressure receptors (Paintal, 1960; Bessou & Laporte, 1961).

The adequate stimulus for the fibres which we have classified as noci-

ceptive is apparently a marked pressure gradient of the sort produced by any sharp object capable of penetrating the integument. Their receptive fields consisted of spot-like areas which, on the posterior thigh, were typically scattered over 10 cm^2. Almost all of the nociceptive fibres had conduction velocities under 30 m/sec and the modal and mean conduction velocities were less than 20 m/sec. While relatively infrequent compared to other receptors such as those associated with hair, they are present in sufficient numbers so as to be readily found in our experiments, representing 15% of the fibres conducting under 30 m/sec.

Cutaneous myelinated fibres range in conduction velocity from under 10 m/sec to over 80 m/sec. Since the nociceptors were concentrated below 30 m/sec, the possibility that fibres of a particular conduction velocity convey information related to a particular cutaneous sensory modality is of considerable interest (see Gasser, 1934). For example, it has recently been argued that as one progresses from large diameter to small diameter across the spectrum of fibres from mechanoreceptors, threshold and resting discharge increase and adaptation is slower (Melzack & Wall, 1965). This idea has been used as the basis for a theory of pain which proposes that certain central cells are released from tonic inhibitory influences by prolonged activity in slowly conducting fibres (Melzack & Wall, 1965). In the present experiments, no appreciable resting activity was observed in any of the fibres conducting below 40 m/sec. The afferent fibres most sensitive to mechanical stimuli in a group of 513 conducting between 6 and 50 m/sec were among the most slowly conducting and rapidly adapting. These, the hair receptors, had a limited conduction velocity range centred at about 18 m/sec and represent the great bulk of afferent fibres conducting under 30 m/sec. The moderate pressure receptors and some of the insensitive mechanoreceptors described above adapted more slowly than hair receptors and typically conducted more rapidly. The most slowly conducting myelinated fibres mainly originated from nociceptors and these adapted relatively quickly to adequate stimulation.

If one considers only the receptors with certain similar characteristics, such as a relative insensitivity to mechanical stimuli and large receptive fields with distinct points of heightened sensitivity, there does appear to be a rough inverse relation between conduction velocity and mechanical sensitivity. We arbitrarily divided our population of 'high-threshold' mechanoreceptors into three groups based upon their response to certain stimulators used in the study. The group labelled nociceptive responded *only* to stimuli which were painful to human subjects and noxious to the cat's skin. A few of these units responded weakly to von Frey hairs exerting forces of more than 3·2 g. A second group of receptors, classified as 'insensitive' mechanoreceptors, seemed similar to the nociceptive group,

except that they were excited by stimuli which were not painful to most human observers, and had von Frey hair thresholds between 0·5 and 3·2 g. The third group, called 'moderate-pressure' receptors, had von Frey thresholds between 0·1 and 0·8 g. On the average, the latter had the most rapidly conducting afferent fibres of the three and differed from the nociceptive group and the 'insensitive 'group in some of their responsive characteristics. Nevertheless, the von Frey hair threshold data indicates that there may, in fact, have been a continuum of thresholds to mechanical stimuli across the three groups. Another consideration is that fibres of each group distinguished between sharp and blunt stimulators in a qualitative (nociceptors) or quantitative sense. At this point, it appears that further understanding of the functional significance of such a threshold continuum in mechanoreceptors awaits knowledge on the way central neural mechanisms are activated by these afferent fibres.

It is possible that receptors with similar receptive fields might differ in receptor morphology and that morphology might correlate with threshold. It is also conceivable that differences in threshold and perhaps in the groups described could reflect differences in the location of undifferentiated terminals. We have no evidence on the structure of the receptors and can only speculate that they may be unencapsulated nerve endings, since this is the most common ending other than hair follicle terminals in mammalian hairy skin (Gilbert, 1929; Pendleton, 1928; Weddell, 1961). Another question related to structure concerns the diameter of the afferent axons. A cross-sectional diameter has not been assigned to fibres herein since there is evidence that the factor which relates conduction velocity to diameter for the slower myelinated fibres is different from that suitable for the fastest ones (Hunt & McIntyre, 1960; Bessou & Perl, 1966), but an accurate value has yet to be determined. If the value Ogden & Miller (1966) give for optic nerve is used, the smallest fibres would be just under 2 μ.

The present analysis demonstrates that more than one kind of sensory information is carried by small-diameter myelinated fibres. Furthermore, in the myelinated spectrum, specific responses to noxious stimuli are limited to certain fine fibres. Therefore, it appears likely that the pain occurring in both man and animals when small myelinated fibres are electrically stimulated is due to activation of fibres associated with nociceptors. It is probable that increases in blood pressure and other spinal reactions in the cat which also are evoked solely by slowly-conducting myelinated fibres (Hagbarth, 1952; Perl, 1957; Fernandez de Molina & Perl, 1965) stem from the nociceptors characterized above.

The nociceptors and other high-threshold receptors studied in our experiments did not respond to noxious heating, strong cooling, or chemical stimuli and did not show any sensitization when subjected to repeated

adequate stimulation. However, pain is initiated by noxious thermal stimuli and chemical substances, and hyperalgesia typically follows injury (Lewis, 1942). Pain has long been related to activity conducted so slowly in the periphery that unmyelinated fibres were implicated (Lewis & Pochin, 1937; Gordon & Whitteridge, 1943). Recent studies of single unmyelinated fibres in carnivores have shown that while many unmyelinated fibres are activated by non-noxious stimuli, some respond only to intense thermal and/or mechanical stimulation (Iggo, 1959, 1960; Iriuchijima & Zotterman, 1960; Hensel, Iggo & Witt, 1960). Thus, it seems reasonable to assume that in cat, as well as in man, unmyelinated fibres are involved in transmitting sensory information concerning noxious stimuli. On the other hand, our results provide a basis for a pain mechanism dependent upon activation of specific receptors with myelinated axons.

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